NJC

LETTER

View Article Online
View Journal | View Issue

Cite this: *New J. Chem.*, 2019, **43**, 556

Received 18th September 2018,
Accepted 29th November 2018

DOI: 10.1039/c8nj04758h

rsc.li/njc

An expeditious synthetic route to proteomimetic foldamers†

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α-Helix proteomimetics such as oligobenzamides have been shown to successfully inhibit a broad array of protein–protein interactions (PPIs) mediated by α-helices. Here we report the synthesis of a protected oligobenzamide intermediate, which can be selectively deprotected and alkylated with desired side chains to rapidly afford a library of α-helix proteomimetics.

Interactions between proteins play a vital role in a large number of cellular processes.[1] α-Helices, which are one of the most common secondary structures in proteins, have been shown to mediate a large number of protein–protein interactions (PPIs) in cells. Hence, any misregulation at the α-helix mediated protein–protein interface might lead to a pathological condition. For example, aberrant α-helix mediated PPIs have been shown to be involved in diseases such as amyloid aggregation,[2] cancer[3] *etc.* Therefore, the modulation of PPIs using small molecules is vital for therapeutic intervention.[4–7] However, identification of a specific small molecule that regulates a protein–protein interface is a difficult task[7,8] due to the lack of well-defined small molecule binding sites on the protein surface. Despite the challenges, several reports have documented the modulation of PPIs using foldamers, which are structural and functional mimics of protein secondary structures.[8–18] A particularly successful approach to targeting helix-mediated PPIs has been to mimic key side chain residues *i*, *i* + 4 and *i* + 7 on one face of the α-helix using proteomimetic foldamers (Fig. 1A).[8,19–23]

Several families of α-helix based proteomimetics have been described by Hamilton and others with potential activity *in vitro* and *in vivo*.[7,13,16,19,24–28] Within this class of modulators, the oligobenzamide scaffold has been used to successfully inhibit an array of α-helix mediated PPIs.[7,13,24,25] A practical limitation of several of these proteomimetics is that each molecule has to be synthesized individually through a series of steps. For the oligobenzamide scaffold, two methods have been largely reported. The first one involves iterative amide coupling on the N-terminal side of the *p*-amino benzoic acid building block, containing the helix mimetic side chain *ortho* to the amino group.[24,29] The second method follows chain elongation on the carboxyl terminal side of the molecule, either by coupling in solution[30] or on a solid phase.[31,32] This process is repeated to synthesize each oligobenzamide molecule. As testing for biological activity against the desired PPI usually requires several such molecules, a library of oligobenzamides needs to be generated which is synthetically demanding.

Herein, we report a new approach for synthesizing an oligobenzamide library. Our method proceeds through a key

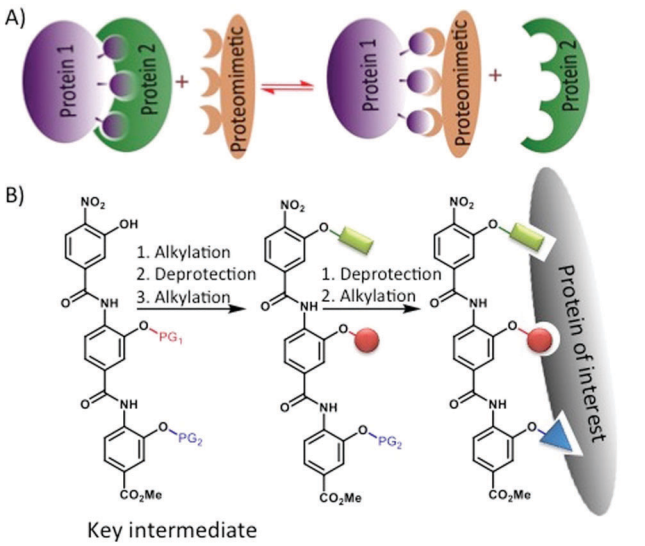

Fig. 1 A strategy for disruption of α-helix mediated PPIs using α-helix proteomimetics. (A) A scheme depicting competitive inhibition of α-helix mediated PPIs by a proteomimetic foldamer (orange) at the interface of two proteins. (B) Facile library generation by subjecting the *o*-protected oligobenzamide intermediate to selective deprotection followed by incorporation of the desired side chains.

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† Electronic supplementary information (ESI) available. See DOI: 10.1039/c8nj04758h
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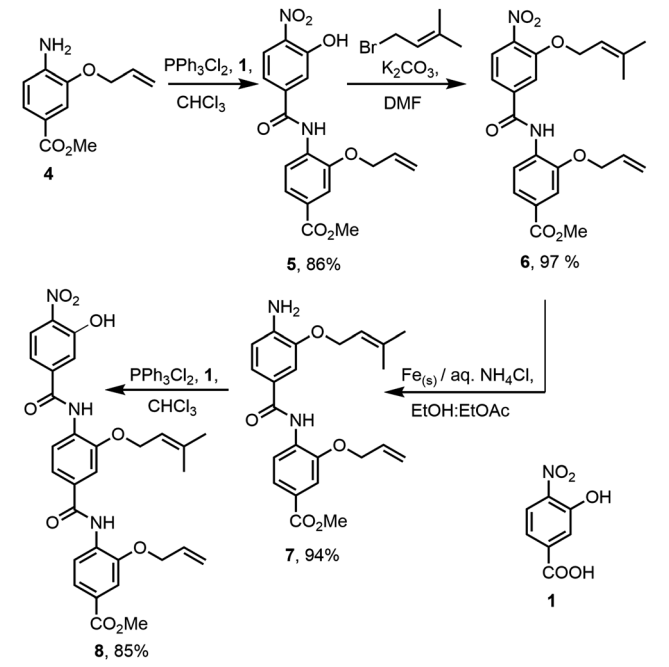
intermediate, which is amenable to late stage modification to generate a library of α-helix mimetics in a facile manner (Fig. 1B). We use selective deprotection of phenolic hydroxy groups followed by alkylation to introduce desired side chains, rapidly generating a oligobenzamide library containing a diverse combination of amino acid mimicking side chains.

We first synthesized a key molecule **8** containing one free hydroxyl group, and two hydroxyl groups protected as ethers. The phenolic ethers, which are inert under our amide coupling conditions, can be selectively cleaved to incorporate the desired side chains as required, yielding final molecules containing helix mimetic side chains at the three phenolic oxygen atoms. This strategy allowed us to generate a library with varied side chain combinations, and indeed made the route expeditious.

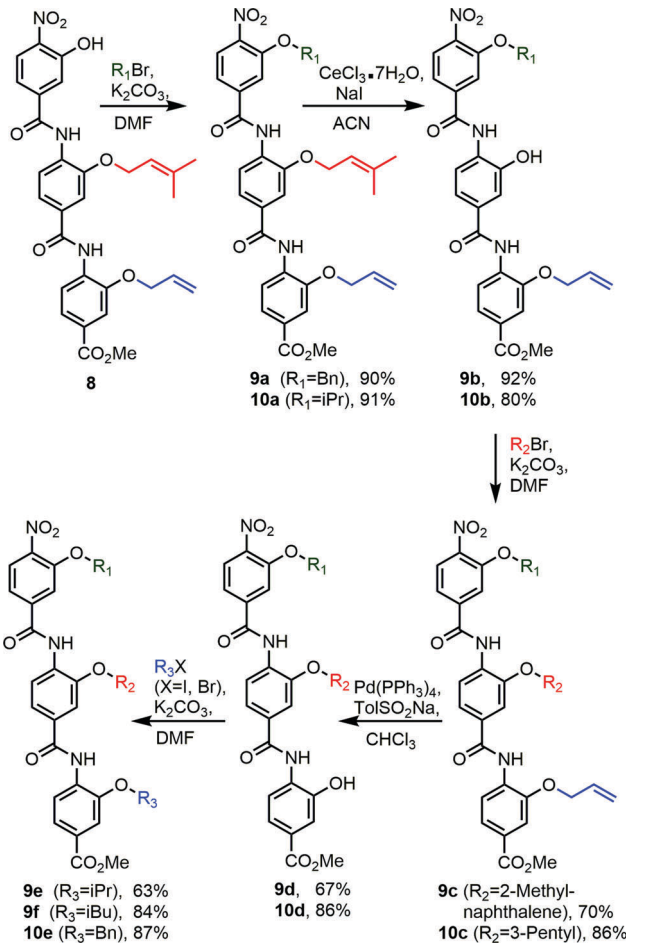
The synthesis (Scheme 1) began with monomer **4** as the building block, synthesized using the reported procedures[30] from **1** *via* intermediates **2–3** (Scheme S1, ESI†). The allyl ether was chosen as the first protecting group for phenol. The allyloxy aniline **4** was coupled to 3-hydroxy-4-nitrobenzoic acid **1** using PPh_3Cl_2 to yield the bisbenzamide **5** with one free hydroxyl group. This reaction is chemoselective and affords 86% conversion to the desired amide. Next, we alkylated the bisbenzamide **5** with prenyl bromide to yield molecule **6**. The prenyl ether serves as the second phenol-protecting group. Reduction of the nitro bisbenzamide **6** by treating with Fe and aq. NH_4Cl yielded the corresponding amino compound **7**, which was coupled with **1** to yield the key oligobenzamide trimer **8**. The molecule **8** was subsequently used as a starting material to generate a library of proteomimetic foldamers by selective deprotection and alkylation of phenolic ethers to afford all possible combinations of side chains. The allyl ether in molecule **8** can be cleaved selectively by treating it with $Pd(PPh_3)_4$ and sodium toluene sulfinate,[30,33] while the prenyl ether can be cleaved selectively with $CeCl_3{\cdot}7H_2O$ and NaI.[33,34]

To generate the library of α-helix mimetic oligobenzamides, we directly subjected molecule **8** to alkylation. In the given example, we have used benzyl or isopropyl groups as representative side chains yielding compounds **9a** and **10a**. Subsequent deprotection of the prenyl group by treating with $CeCl_3{\cdot}7H_2O$ and NaI yielded **9b** and **10b**. Under the reaction conditions, the cleavage was selective for the prenyl ether, leaving the allyl ether intact. The alkylation and arylation of **9b** and **10b** yielded molecules **9c** and **10c** respectively, each containing two α-helix mimetic side chains. The deallylated compounds **9d** and **10d** obtained from **9c** and **10c** were subjected to the third *O*-alkylation by reacting with the desired bromo derivative to yield the final compounds **9e**, **9f**, and **10e** (Scheme 2). This sequential deprotection and alkylation scheme allowed us to generate helix mimetics with three different side chains *i.e.* $R_1 \neq R_2 \neq R_3$. Each of the above steps proceeded in good to excellent yields giving 24–47% overall yield over five steps for the final molecules containing non-identical side chains starting from **8** (Table 1).

In cases where the same side chain is desired at more than one of the three positions, the above procedure can be further



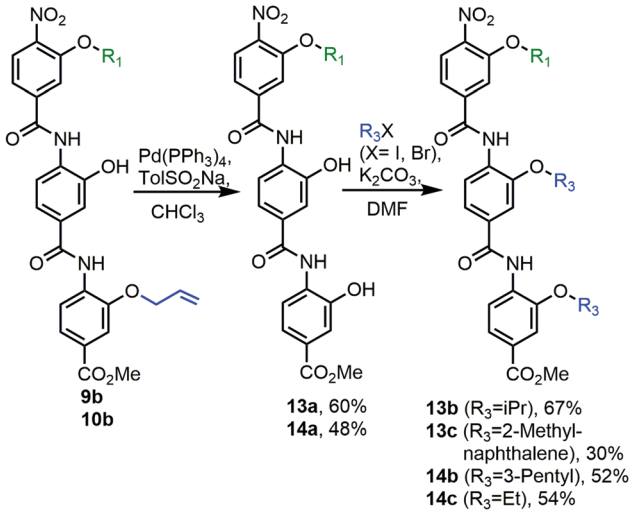
Scheme 1 Synthesis of the key protected oligobenzamide intermediate.



Scheme 2 Generation of proteomimetics with three non-identical side chains ($R_1 \neq R_2 \neq R_3$).

Table 1 Library of molecules synthesized using the selective deprotection and alkylation route

Molecule	R_1	R_2	R_3	Overall yield starting from **8** (%)
9e	Bn	2-Methylnaphthalene	^{i}Pr	24
9f	Bn	2-Methylnaphthalene	^{i}Bu	33
10e	^{i}Pr	3-Pentyl	Bn	47
11c	Bn	Bn	3-Pentyl	22
12c	^{i}Pr	3-Pentyl	^{i}Pr	32
12d	^{i}Pr	Bn	^{i}Pr	36
13b	Bn	^{i}Pr	^{i}Pr	33
13c	Bn	2-Methylnaphthalene	2-Methylnaphthalene	15
14b	^{i}Pr	3-Pentyl	3-Pentyl	18
14c	^{i}Pr	Et	Et	19
15a	^{i}Pr	^{i}Pr	^{i}Pr	33
15b	Bn	Bn	Bn	34
16a	^{i}Pr	^{i}Pentyl	Pr	57
16b	^{i}Pr	3-Pentyl	Pr	47
16c	^{i}Pr	^{i}Pentyl	^{i}Pr	45



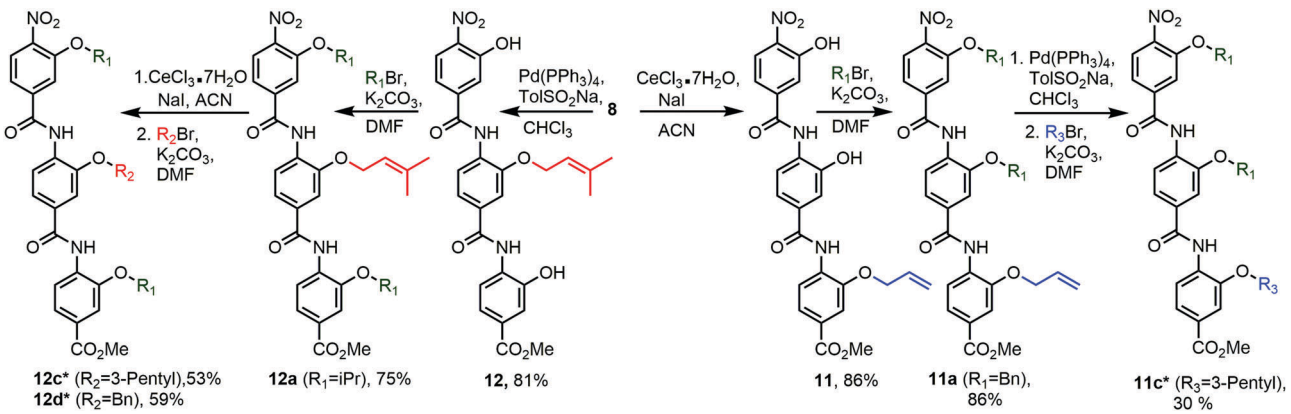
Scheme 4 Generation of proteomimetics with two identical side chains ($R_1 \neq R_2 = R_3$).

stream-lined making it more efficient. As an example, we deprotected the prenyl group in **8** to afford molecule **11** (Scheme 3, right). Alkylation of molecule **11** yielded molecules with two identical side chains at the top. This has been demonstrated with benzylation (**11a**). The deallylation of **11a** followed by alkylation using 3-bromopentane afforded molecule **11c** (Scheme 3, right). This strategy can be used for generating a second combination *i.e.* $R_1 = R_2 \neq R_3$, where two identical –R groups can be incorporated simultaneously. The third combination $R_1 = R_3 \neq R_2$ was synthesized similarly by deprotecting the allyl ether[30] in intermediate **8** to afford compound **12** leaving the prenyl ether intact. The alkylation of the two phenolic hydroxy groups with desired alkyl halides such as 2-bromopropane resulted in compound **12a**. Subsequent removal of the prenyl group and further alkylation with 3-bromopentane or benzyl bromide resulted in the final molecules **12c** and **12d** (Scheme 3, left).

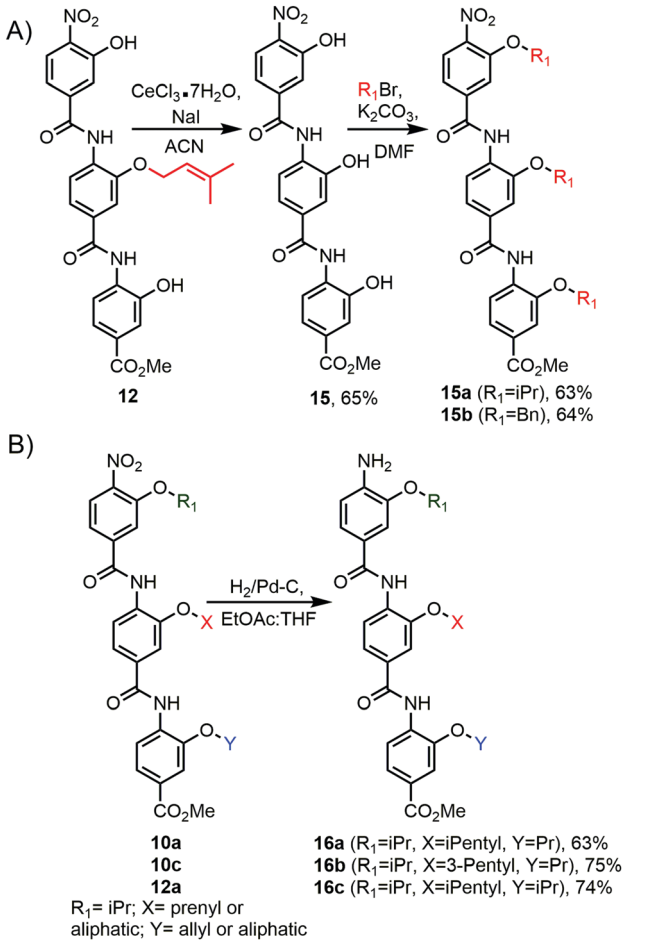
To yield the fourth combination $R_1 \neq R_2 = R_3$, we directly deallylated the intermediates **9b** and **10b** to yield molecules **13a** and **14a**. Further, dialkylation with desired alkyl halides such as 2-bromo propane, 2-bromo(methyl)naphthalene, 3-bromopentane and iodoethane yielded the corresponding molecules **13b**, **13c**, **14b** and **14c** (Scheme 4).

In cases where the same side chain was required in all positions, the prenyl ether from compound **12** was removed to yield the fully deprotected oligobenzamide **15**. Alkylation of **15** with the desired bromide substrate readily resulted in molecules where all three side chains were identical *i.e.* $R_1 = R_2 = R_3$ (Scheme 5A). To generate additional diversity, we subjected the intermediate molecule bearing the prenyl and allyl groups **10a**, **10c** and **12a** to hydrogenation to yield molecules **16a**, **16b** and **16c** (Scheme 5B and Table 1). Using the deprotection and alkylation strategy described above, we synthesized a representative library of molecules listed in Table 1, which will be used to screen against α-helix mediated PPIs.

In summary, we have developed a new synthetic approach to synthesize a library of *o*-alkoxy oligobenzamides, which act as α-helix mimetics. Our approach involves the selective deprotection and alkylation of a key intermediate to readily generate all possible combinations of side chains, thus making the route



Scheme 3 Generation of proteomimetics with two identical side chains. The right route affords combination $R_1 = R_2 \neq R_3$ whereas the left route affords combination $R_1 = R_3 \neq R_2$ (* yields are over two steps).



Scheme 5 (A) Generation of proteomimetics with three identical side chains (R_1 = R_2 = R_3). (B) Reduction of nitro, allyl and prenyl groups in one step to generate additional diversity.

simple and efficient. The protection and deprotection strategy described here can be readily applied to other scaffolds for their selective functionalization. In future, the activity of the synthesized molecules will be tested against α-helix mediated protein–protein interfaces.

Conflicts of interest

There are no conflicts to declare.

Acknowledgements

We thank SERB and IISER Bhopal for financial support. BAB thanks CSIR for a Senior Research Fellowship. IS is a recipient of the Ramanujan fellowship. The authors thank the Saraogi lab members for valuable suggestions.

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